Characterization of Human Coronavirus OC43 and Human Coronavirus NL63 Infections Among Hospitalized Children <5 Years of Age

Jina Lee, MD, PhD†* and *Gregory A. Storch, MD**

Background: Multiplex molecular assays now make it possible for clinical laboratories to detect human coronaviruses (HCoVs). We investigated the clinical characteristics of HCoV-OC43 and HCoV-NL63 in patients <5 years of age during a recent coronavirus season.

Methods: Respiratory viruses were detected using a multiplex molecular assay at St. Louis Children`s Hospital starting in November 2012. We analyzed demographic and clinical data from all patients <5 years of age with solo detection of HCoV-OC43 (n = 52) and HCoV-NL63 (n = 44) and for comparison, samples of children with respiratory syncytial virus, parainfluenza virus and picornaviruses.

Results: During the study period, HCoV-OC43 (4%) was the 5th and HCoV-NL63 the 8th (2%) most common respiratory virus. Coinfections were detected in 35% and 38% of children with HCoV-OC43 and HCoV-NL63, respectively. Croup was more common with HCoV-NL63 (30%) than with HCoV-OC43 (2%). Lower respiratory tract infection occurred in 33% of children with HCoV-OC43 and 25% of children with HCoV-NL63. Severe illness was less common in HCoV-NL63, HCoV-OC43 and parainfluenza virus (14%, each) compared with respiratory syncytial virus (30%) and picornaviruses (26%; $P = 0.055$ for HCoVs combined compared with the other respiratory viruses) and occurred mainly in those with underlying medical conditions.

Conclusions: Infections caused by HCoV-OC43 and HCoV-NL63 are common and include some with lower respiratory tract involvement and severe disease, especially in children with underlying medical conditions. Overall, a substantial burden of disease associated with both HCoV-OC43 and HCoV-NL63 was observed for hospitalized children <5 years of age.

Key Words: coronavirus, HCoV-OC43, HCoV-NL63, children, outbreak, clinical severity, respiratory viruses

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Since human coronaviruses (HCoVs) were initially discovered in the 1960s, 6 viruses including HCoV-229E, HCoV-OC43, HCoV-NL63, HCoV-HKU1, severe acute respiratory syndrome (SARS)-CoV and Middle East Respiratory Syndrome-CoV have been recognized as the causative agents of a range of respiratory tract infections. Epidemiological surveys have shown that HCoVs are worldwide in distribution and circulate during seasonal outbreaks. Among the HCoVs, HCoV-OC43 is the most common.[1] HCoV-NL63 and HCoV-HKU1 were described in 2004 and 2005, respectively, even though they had circulated in humans for many years.[2] SARS-CoV was associated with an outbreak of SARS in 2003. In 2012, the new highly virulent Middle East Respiratory Syndrome-CoV emerged in the Middle East and showed pathogenicity similar to SARS-CoV with a high fatality rate.[3] Except for SARS-CoV and Middle East Respiratory Syndrome-CoV, the HCoVs have been regarded as mild pathogens. An exception was a novel genotype of HCoV-OC43 resulting from recombination that was identified as a cause of more severe disease in elderly patients.[4]

In November 2012, the St. Louis Children's Hospital (SLCH) Virology Laboratory implemented a multiplex molecular test for routine detection of respiratory viruses that included the capability to detect all 4 common HCoVs. With the high volume of respiratory virus testing performed in the laboratory, large numbers of patients positive for HCoVs were identified, most of which were HCoV-OC43 or HCoV-NL63. Because of uncertainty regarding how to interpret the detection of these viruses in the routine diagnostic setting, we undertook a study to determine the clinical findings and significance of HCoV-OC43 and HCoV-NL63 in children who were admitted to the hospital or visited the emergency department (ED). We compared findings in these children to those in children infected with other commonly detected respiratory viruses including human rhinovirus/enterovirus (RV/EV), respiratory syncytial virus (RSV) and parainfluenza viruses (PIV) who were also hospitalized or seen in the ED during the same period.

MATERIALS AND METHODS

Epidemiologic Analysis

Data from the Virology Laboratory at SLCH was used to determine the relative frequency of respiratory viruses detected at SLCH from November 2012 to November 2013.

Clinical Analysis

Clinical and demographic data from all patients <5 years of age with single infections of HCoV-OC43 or HCoV-NL63 who were hospitalized or visited the ED at SLCH from November 2012 to March 2013 were abstracted from electronic medical records. To include a comparable number of HCoV-NL63 cases, all patients <5 years of age with HCoV-NL63 infection, which was detected from April 2013 to July 2013, were also included in the analysis. Clinical information including diagnosis, hospitalization, underlying medical conditions, intensive care unit stay and need for mechanical ventilation was examined. Mechanical ventilation included both invasive ventilation requiring tracheal intubation and noninvasive ventilation such as continuous positive airway pressure and bilevel positive airway pressure. Only the first virus detected during a single clinical episode occurring within a 4-week period was included in the analysis, and duplicates from the same patient were excluded. We also abstracted information from the medical

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records of patients <5 years of age with single infections of RV/EV, RSV and PIV. For each of these viruses, we reviewed the records of a comparable number of patients with each of these viruses who were randomly selected from lists of all patients <5 years of age, who were positive for the virus being analyzed from November 2012 to March 2013.

Case Definitions

Lower respiratory tract infection (LRTI) was defined as bronchiolitis or pneumonia. Bronchiolitis was diagnosed based on the presence of signs of lower airway obstruction, such as wheezing, retraction and decreased breath sounds, and either a normal chest radiograph or a chest radiograph showing hyperinflation or mild perihilar infiltrates. No effort was made to distinguish between asthma exacerbation and bronchiolitis. Pneumonia was diagnosed in patients with characteristic adventitious sounds such as crackles and a chest radiograph with local infiltrates or consolidation. Upper respiratory tract infection was defined as coryza, nasal obstruction, cough or pharyngitis without manifestations of lower respiratory tract disease. If no symptoms or signs were observed suggestive of infection of any system, the virus was considered to be an innocent bystander. An episode resulting in intensive care unit stay, mechanical ventilation and/or death was considered to be severe disease.

Microbiological Testing

With the exception of specimens submitted only for rapid influenza or RSV testing, specimens submitted to the Virology Laboratory at SLCH for respiratory virus detection were tested using the FilmArray Respiratory Panel (BioFire Diagnostics, Salt Lake City, UT), which is a multiplexed nucleic acid test capable of simultaneous qualitative detection and identification of multiple respiratory viruses and bacteria. The following viruses, virus subtypes and bacteria could be identified using the FilmArray Respiratory Panel: adenovirus, HCoV-HKU1, HCoV-NL63, HCoV-229E, HCoV-OC43, influenza A, influenza A subtypes H1, H3 and 2009 H1, influenza B, human metapneumovirus, PIV types 1, 2, 3 and 4, RSV, RV/EV, *Bordetella pertussis*, *Chlamydophila pneumoniae* and *Mycoplasma pneumoniae*. Because of the genetic similarity between human RV and EV, the FilmArray Respiratory Panel cannot reliably differentiate them, and they were grouped as picornaviruses in this study.

Statistical Analysis

Pearson χ^2 test and analysis of variation were used to compare categorical and continuous variables, respectively. Multiple logistic regression analysis was used to determine whether each respiratory viral infection was independently associated with clinical severity. All analyses were performed using SPSS version 13.0 (SPSS, Chicago, IL), and the level of statistical significance was set at 0.05.

RESULTS

Prevalence of Respiratory Viruses

The population under investigation was all patients that were tested for the respiratory viruses during the consecutive 13 months from November 2012 to November 2013. The overall positive rate for respiratory viruses was 47%; 4562 of 9628 respiratory samples submitted to the Virology Laboratory at SLCH. The clinical samples were mostly nasopharyngeal swabs (93%) followed by bronchoalveolar lavage fluid (4%), tracheal aspirates (2%) and sputum (1%). The relative frequency of viruses by month is shown in Figure (Supplemental Digital Content 1, http://links.lww.com/INF/B826). The peak incidence of HCoV-OC43 occurred in December 2012 and declined to a low level by March 2013. HCoV-NL63 was first detected in December 2012, peaked in February 2013 and ceased being detected in May 2013.

Of the 4315 viruses detected from patients <18 years of age during the period November 2012 to November 2013, picornaviruses were the most frequent (53%), HCoV-OC43 the 6th (4%) and HCoV-NL63 the 8th (2%; Table 1). Among the positive samples, coinfection with ≥2 viruses was present in 862 (20%). No patient had >3 different respiratory viruses at once. Coinfection was more likely to include HCoV-OC43 (35%) and HCoV-NL63 (38%), compared with RSV (23%), picornaviruses (14%) and PIV (26%; $P < 0.001$ for HCoVs combined compared with all of the other viruses; Table 1).

TABLE 1. Viruses Detected in 4315 Respiratory Samples from Patients <18 Years of Age at St. Louis Children´s Hospital, from November 2012 through November 2013

Virus	Single Virus Detected	Indicated Virus With 1 Other Virus	Indicated Virus with 2 Other Viruses*	Total Number of Patients With the Indicated Virus	Percent of Total†	Coinfection (%)‡
Ad	131	74	5	210	5	38
HCoV-NL63	62	35	3	100	2	38
HCoV-HKU1	10	10	2	22	1	55
HCoV-229E	8	9	0	17	0	53
HCoV-OC43	102	46	8	156	4	35
IF A	67	7	9	83	2	19
IF B	111	14	4	129	3	14
MPV	186	51	2	239	6	22
PIV	363	121	7	491	12	26
Type 1	149	31	2	182	38	18
Type 2	20	6	0	26	5	23
Type 3	112	51	2	165	34	32
Type 4	80	32	3	115	24	30
Type not determined	2	1	0	3	1	33
Picornavirus (RV/EV)	1928	295	17	2240	53	14
RSV	485	133	10	628	15	23
Total	3453	795	67	4315	103	20

*No patients had >3 different respiratory viruses at once.

†Patients with the indicated virus as a percentage of total number of viruses detected.

‡Number of patients with >1 virus detected as a percent of the total with the indicated virus.

Ad, Adenovirus; IF A, influenza A; IF B, influenza B; MPV, metapneumovirus; RV/EV, rhinovirus/enterovirus (the test does not distinguish between RV and EV).

Demographic Characteristics

Among 2940 patients <18 year of age infected with a single respiratory virus during the period November 2012 to November 2013, 70% were <5 years old and the age distribution of children with each virus is shown in Figure 1. The percentage of patients <5 years of age varied from 64% for HCoV-OC43 to 88% for RSV.

The 254 patients <5 years of age whose records were reviewed included 52 with HCoV-OC43, 46 with HCoV-NL63, 50 with PIV, 54 with picornaviruses and 54 with RSV. Of these patients, 54% were male, 58% were Caucasian and 35% were African American. These and other demographic characteristics were not significantly different among the 5 virus groups (Table 2). Underlying conditions were present more frequently in patients infected with HCoV-OC43 (77%) but not with HCoV-NL63 (48%) compared with children infected with the other respiratory viruses (PIV, 32%; picornaviruses, 67% and RSV, 46%; $P = 0.022$ for HCoVs combined vs. all of the noncoronaviruses). For HCoV-OC43, congenital heart disease was especially common, whereas for picornaviruses and RSV, reactive airways disease/asthma was the most common underlying condition, being present more frequently with picornavirus and RSV than with the other viruses ($P = 0.002$).

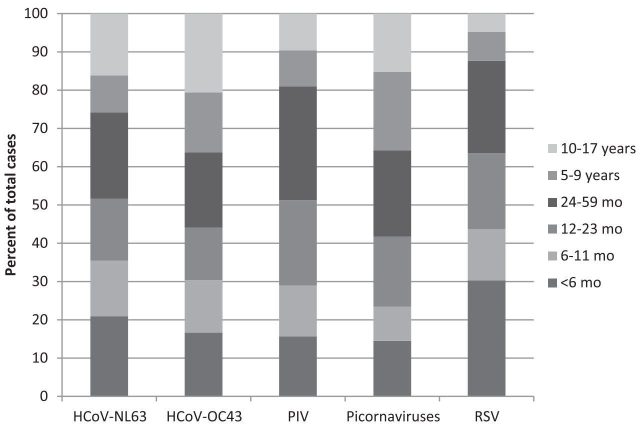

FIGURE 1. Age distribution of 2940 children <18 years of age infected with a single respiratory virus from November 2012 to November 2013. The percentage of the positives who were <5 years old are as follows: HCoV-NL63, 74%; HCoV-OC43, 64%; PIV, 81%; picornavirus, 64% and RSV, 88% ($P < 0.001$).

Clinical Characteristics

The most common diagnosis in young children with HCoV-OC43 was upper respiratory tract infection (54%), followed by LRTI (33%) and acute otitis media (19%; Figures, Supplemental Digital Content 2 and 3, http://links.lww.com/INF/B827 and http://links.lww.com/INF/B828). In comparison, croup (30%) was the most frequent diagnosis in children with HCoV-NL63 and was significantly more common in children with HCoV-NL63 than in those with HCoV-OC43 (2%; $P < 0.001$) or any of the other respiratory viruses (PIV, 16%; picornaviruses, 6% and RSV, 4%; $P < 0.001$ for HCoV-NL63 compared with all of the noncoronaviruses). LRTI was less common in children with HCoV-OC43 (33%) and HCoV-NL63 (25%) compared with those with RSV (83%), picornavirus (59%) and PIV (38%; $P < 0.001$ for incidence of LRTI for HCoVs combined compared with noncoronaviruses). Approximately 20% of children infected with HCoV-OC43, RSV and PIV were diagnosed with acute otitis media, compared with only 5% of those with HCoV-NL63. Wheezing was less frequently observed in children with HCoVs compared with PIV, RSV and picornavirus ($P < 0.001$ for HCoVs combined compared with all of the noncoronaviruses; Table 3).

Clinical Diagnoses According to Age

All of the croup cases among the patients <6 months of age were associated with HCoV-NL63 infection although HCoV-NL63-associated croup was also observed throughout the

TABLE 2. Demographic Characteristics of Patients < 5 Years of of Age Infected With a Single Respiratory Virus

	Single Virus Detected					
	HCoV-NL63 (n = 44)	HCoV-OC43 (n = 52)	PIV (n = 50)	Picornaviruses (n = 54)	RSV (n = 54)	*P* Value *
Mean age in months (SD)	18 (16)	18 (17)	21 (18)	21 (16)	18 (15)	0.765
Race						0.812
African American	17 (39)†	20 (39)	17 (34)	21 (39)	14 (26)	
Caucasian	25 (57)	29 (56)	28 (56)	29 (54)	37 (69)	
Other	2 (5)	3 (6)	5 (10)	4 (7)	3 (6)	
Male gender	20 (46)	27 (52)	31 (62)	34 (63)	25 (46)	0.222
Nosocomial acquisition	0 (0)	1 (2)	2 (4)	2 (4)	1 (2)	0.661
Hospitalized	35 (80)	45 (87)	38 (76)	48 (89)	50 (93)	0.116
Known sick contact	13 (30)	12 (23)	18 (36)	11 (20)	28 (52)	0.003
Attending DCC	7 (16)	10 (19)	8 (16)	5 (9)	14 (26)	0.143
Underlying disease						
Asthma/RAD	7 (16)	4 (8)	3 (6)	17 (32)	13 (24)	0.002
Congenital heart disease	5 (11)	14 (27)	2 (4)	2 (4)	2 (4)	<0.001
Prematurity (GA ≤34 wks)	4 (9)	8 (15)	4 (8)	7 (13)	8 (15)	0.720
Chronic respiratory condition‡	5 (11)	6 (12)	3 (6)	8 (13)	7 (13)	0.700
Immunosuppressed§	4 (9)	3 (6)	2 (4)	4 (7)	3 (6)	0.875
Neuromuscular disease	6 (14)	13 (25)	4 (8)	10 (19)	5 (9)	0.092
Sickle cell disease	1 (2)	5 (10)	1 (2)	2 (4)	0 (0)	0.086
Other¶	6 (14)	13 (25)	3 (6)	7 (13)	4 (7)	0.036
None	23 (52)	12 (23)	34 (68)	18 (33)	29 (54)	<0.001

*P value was calculated by analysis of variation for comparison of mean age according to each virus group and by Pearson χ^2 test for categorical variables.
†Parenthesis shows percent in each indicated case.
‡Cystic fibrosis, upper airway problems, bronchopulmonary dysplasia and tracheostomy, does not include asthma or recurrent wheezing.
§ Malignancy, hematopoietic stem cell transplantation, solid organ transplantation and corticosteroid treatment.
¶Genetic disorders or syndromes, failure to thrive, gastrointestinal problems and chronic liver disease.
SD, standard deviation; DCC, day care center; RAD, reactive airway disease; GA, gestational age.

TABLE 3. Symptoms and Signs Among Patients <5 Years of Age Infected With a Single Respiratory Virus

	Single Virus Detected					
	HCoV-NL63 (n = 44)	HCoV-OC43 (n = 52)	PIV (n = 50)	Picornaviruses (n = 54)	RSV (n = 54)	*P* Value*
	Number (%)					
Symptoms and signs						
Fever (≥38.0°C)	20 (46)	29 (56)	31 (62)	21 (39)	36 (67)	0.002
Stridor	13 (30)	2 (4)	9 (18)	4 (7)	5 (9)	0.007
Retractions	18 (41)	14 (27)	16 (32)	28 (52)	37 (69)	0.001
Rales	1 (2)	5 (10)	5 (10)	11 (20)	18 (33)	0.001
Wheezing	8 (18)	10 (19)	16 (32)	28 (52)	31 (57)	<0.001
Decreased breathing sounds	2 (5)	1 (2)	6 (12)	13 (24)	15 (28)	<0.001
Dyspnea	14 (32)	12 (23)	17 (34)	27 (50)	38 (70)	<0.001
Pleural effusion	1 (2)	1 (2)	0 (0)	0 (0)	0 (0)	0.450
Cough	32 (73)	32 (62)	38 (76)	36 (67)	53 (98)	0.003
Sputum	2 (5)	1 (2)	8 (16)	10 (19)	11 (20)	0.006
Rhinorrhea	23 (52)	33 (64)	32 (64)	39 (72)	38 (70)	0.487
Nasal congestion	14 (32)	19 (37)	27 (54)	29 (54)	36 (67)	0.006
Rash	1 (2)	2 (4)	2 (4)	6 (11)	6 (11)	0.157
Vomiting	11 (25)	12 (23)	15 (30	18 (33)	22 (41)	0.235
Diarrhea	8 (18)	5 (10)	4 (8)	7 (13)	6 (11)	0.537
Poor oral intake	13 (30)	17 (33)	16 (32)	21 (39)	37 (69)	0.001
Conjunctival injection	1 (2)	3 (6)	2 (4)	1 (2)	1 (2)	0.653
Hypotension	0 (0.0)	2 (4)	1 (2)	1 (2)	0 (0)	0.586
Lethargy	9 (21)	14 (27)	11 (22)	16 (30)	19 (35)	0.793
Irritability	10 (23)	15 (29)	16 (32)	24 (44)	19 (352)	0.190
Chest radiography						0.001
Normal	7 (16)	20 (38.5)	13 (26.0)	15 (27.8)	12 (22.2)	
Abnormal	12 (27)	16 (30.8)	17 (34.0)	17 (31.5)	32 (59.3)	
Not done	25 (57)	16 (30.8)	20 (40.0)	22 (40.7)	10 (18.5)	

**P* value was calculated by Pearson χ^2 test.

age range up to 5 years. (Fig., Supplemental Digital Content 2, http://links.lww.com/INF/B827). In comparison, the peak incidence of PIV-associated croup was in the 6- to 23-month-old group. In the age group <6 months of age, none of the HCoV-NL63 cases presented with LRTI. This was different from RSV because most of the children in this age group with RSV did present with LRTI.

Clinical Outcome and Severity

Supplemental oxygen therapy was required less frequently in children with HCoV-OC43 (25%) and HCoV-NL63 (18%) compared with the other respiratory viruses (41%; $P < 0.001$) and was most frequent in children with RSV (67%; Table 4). Forty-three percent of children received antibiotics, with no statistically significant differences in children with any of the specific viruses ($P = 0.062$). Overall, 20% (50/254) of cases were classified as severe, with no fatalities. The occurrence of severe disease including intensive care unit admission and mechanical ventilation ranged from 30% for children with RSV and 26% for those with picornaviruses to 14% for those with HCoV-OC43, HCoV-NL63 and PIV, each ($P = 0.055$ for HCoVs combined compared with the other respiratory viruses).

Effect of Underlying Medical Conditions

The presence of an underlying medical condition affected the distribution of respiratory tract diagnoses (Fig., Supplemental Digital Content 3, http://links.lww.com/INF/B828) and the frequency of severe disease (Fig. 2). LRTI associated with both HCoV-OC43 and HCoV-NL63 occurred disproportionately in children with underlying medical conditions [41% (25/61) in children with any underlying medical condition compared with 9% (3/35) in otherwise healthy children; $P = 0.001$]. In contrast, croup associated with HCoV-NL63 occurred with the same relative frequency, regardless of the presence of an underlying medical condition [26% (6/23) in previously healthy children compared with 33% (7/21) in children with underlying medical conditions; $P = 0.599$]

Severe disease in children with HCoV-OC43 and HCoV-NL63 occurred more often in children with than in those without underlying medical conditions (Fig. 2). In children with underlying conditions, the percentage of children with severe disease was similar for each of the viruses ($P = 0.827$). In contrast, in children without underlying conditions, RSV and picornaviruses were more likely and HCoV-OC43 and HCoV-NL63 were less likely than the other viruses to be associated with severe disease ($P = 0.026$).

We used univariate and multivariate logistic regression to further analyze risk factors for severe disease. In the univariate analysis, the presence of congenital heart disease and a chronic respiratory condition were significant risk factors for severe disease (Table 5). No specific virus was a risk factor in the univariate analysis. With adjustment for age and the underlying medical conditions of congenital heart disease and chronic respiratory conditions in a multiple logistic regression analysis, RSV and no other virus were significantly associated with severe disease (adjusted odds ratio, 3.4; 95% confidence interval: 1.1–10.4; Table 5).

DISCUSSION

Our study of the clinical manifestations of HCoV infections encountered in a hospital diagnostic virology laboratory was based on the recent implementation of multiplex molecular testing for respiratory pathogens, which provided the capability to detect HCoV infections that had not been available previously in most diagnostic virology laboratories.

As reported by others,[5,6] 2 HCoVs, HCoV-OC43 and HCoV-NL63 were common, being detected in 4% and 2%,

TABLE 4. Clinical Outcomes and Severity in Children <5 Years of Age Infected With A Single Respiratory Virus

	Virus					
	HCoV-NL63 (n = 44)	HCoV-OC43 (n = 52)	PIV (n = 50)	Picornaviruses (n = 54)	RSV (n = 54)	*P* Value*
Intervention/Severity	Number (%)					
Antibiotic use	15 (34)	26 (50)	22 (44)	16 (30)	29 (54)	0.062
Parenteral	13 (30)	21 (40)	15 (30)	15 (28)	21 (39)	0.527
Per oral	8 (18)	14 (27)	12 (24)	9 (17)	19 (35)	0.181
Nebulizer use	23 (52)	18 (35)	20 (40)	34 (63)	36 (67)	0.010
Systemic steroid use	13 (30)	5 (10)	11 (22)	23 (43)	24 (44)	<0.001
Supplementary O_2 need	8 (18)	13 (25)	13 (26)	25 (46)	36 (67)	<0.001
Severe disease†	6 (4)	7 (14)	7 (14)	14 (26)	16 (30)	0.091
ICU stay	6 (14)	7 (14)	6 (12)	13 (24)	16 (30)	0.080
Invasive MV use‡	0 (0.0)	4 (8)	1 (2)	2 (4)	2 (4)	0.332
Noninvasive MV use§	2 (5)	3 (6)	2 (4)	4 (7)	4 (7)	0.923

**P* value was calculated by Pearson χ^2 test.
†An episode resulting in ICU stay, mechanical ventilation and/or death was considered a severe case.
‡Invasive MV indicates any kind of mechanical ventilator requiring tracheal intubation.
§Noninvasive MV indicates continuous positive airway pressure and bilevel positive airway pressure
ICU, intensive care unit; MV, mechanical ventilator.

respectively, of specimens from children <18 years of age. Also as reported previously, both viruses were frequently present along with 1 or more other respiratory viruses, with a 36% rate of coinfection. Although the clinical impact of coinfection was not evaluated in this study, some studies suggested the presence of coinfection may not affect the clinical severity and characteristics of respiratory tract infection.[7]

To discern the clinical manifestations associated with each virus, we focused our attention on cases in which HCoVs were the only viruses detected. We found that children infected with HCoVs had milder illness than children infected with RSV and picornaviruses. As seen in other studies,[7,8] most of the cases of HCoV-OC43 were in children with underlying medical conditions, also suggesting that these infections are relatively mild. However, a minority of children with either HCoV-OC43 and HCoV-NL63 had lower respiratory tract involvement and/or had severe disease requiring mechanical ventilation and intensive care. Most of the HCoV-OC43 cases occurred in children with underlying diseases, in contrast to RSV, which was often associated with severe disease in otherwise normal children.

We found several other distinctive clinical patterns, some of which have been noted previously.[9–11] Most notably, HCoV-NL63 but not HCoV-OC43 was highly associated with croup, even more so than PIV, and appeared to be the predominant cause of croup in young infants <6 months of age. In contrast, as noted by others,[6,12] we found an association between HCoV-OC43 and otitis media, with one-fifth of the HCoV-OC43-infected children being diagnosed with acute otitis media. A similar association was not apparent for HCoV-NL63. We are not aware of an explanation for the association of HCoV-OC43 and otitis media.

A recent prospective longitudinal cohort study of previously healthy young children showed that HCoVs were detected in 8% (10/119) of children with LRTI, but were not detected in any children with LRTI who were ≥2 years of age.[6] In our study, however, HCoV-OC43 and HCoV-NL63 were associated with LRTIs in children ≥2 years of age (29%, 9/31) as well as in those <2 years of age (29%, 19/65). The greatest incidence of LRTIs associated with HCoVs combined was between 6 and 11 months of age, which was older than the peak age of <6 months for RSV- and PIVs-associated LRTIs but younger than that of 12–23 months with picornavirus-associated LRTIs. These findings may relate to the difference in decline of maternal antibody titers or in the degree of immunoprotection for each virus.[6] In one study, high levels of antibodies to HCoVs-OC43, HCoVs-NL63, HCoVs-HKU1 and HCoVs-229E were observed at birth and decreased to low detectable levels within a few months.[13]

This study has several strengths and limitations. A strength is that it was based on testing performed in a routine US hospital diagnostic laboratory and, to our best knowledge, included more children infected only with HCoVs compared with any previous

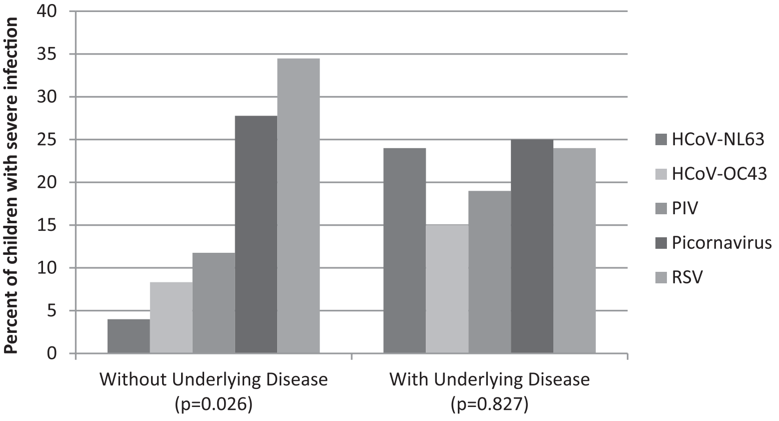

FIGURE 2. The percentage of patients <5 years of age infected with respiratory viruses who had severe infections, according to the presence or absence of an underlying diseases. In children with underlying conditions, the percentage of children with severe disease was similar for each of the viruses (*P* = 0.827). In contrast, in children without underlying conditions, RSV and picornaviruses were more likely and HCoV-OC43 and HCoV-NL63 were less likely than the other viruses to be associated with severe disease (*P* = 0.026).

TABLE 5. Logistic Regression Analysis of Risk Factors for Severe Disease Associated With Respiratory Viral Infections

Variable	OR (95% CI) From Univariate Analysis	Adjusted OR (95% CI) From Multivariate Analysis
Age	1.0 (1.0–1.0)	1.0 (1.0–1.0)
Race		NA
Caucasian	1.0*	
African American	0.9 (0.4–1.7)	
Other	1.2 (0.4–4.0)	
Underlying medical condition		
None	0.8 (0.4–1.6)	NA
Congenital heart disease	2.6 (1.1–6.2)	4.7 (1.6–13.6)
Prematurity (GA ≤34 wks)	1.0 (0.4–2.5)	NA
Chronic respiratory condition†	3.5 (1.5–7.9)	3.5 (1.5–8.3)
Asthma/RAD	0.7 (0.3–1.8)	NA
Immunodeficiency‡	0.3 (0.0–2.0)	NA
Neuromuscular disease	1.3 (0.6–3.0)	NA
Sickle cell disease	0 (0–∞)	NA
Other§	1.6 (0.7–3.8)	NA
Viruses		
HCoV-NL63	1.0*	1.0*
HCoV-OC43	1.0 (0.3–3.2)	0.7 (0.2–2.5)
PIV	1.0 (0.3–3.3)	1.5 (0.4–5.1)
Picornaviruses	2.2 (0.8–6.4)	2.9 (0.9–8.8)
RSV	2.7 (0.9–7.5)	3.4 (1.1–10.4)

*Reference value.

†Cystic fibrosis, upper airway problems, bronchopulmonary dysplasia and tracheostomy, does not include asthma or recurrent wheezing.

‡Malignancy, hematopoietic stem cell transplantation, solid organ transplantation and corticosteroid treatment.

§Genetic disorders or syndromes, failure to thrive, gastrointestinal problems and chronic liver disease.

OR, odds ratio; CI, confidence interval; GA, gestational age; RAD, reactive airway disease; NA, not applicable.

study. Also, it included comparison groups of children infected with 1 of 3 well-known viruses: RSV, PIV and picornaviruses. A limitation is that it did not include asymptomatic children. Several previous studies have shown similar rates of detection of HCoVs in asymptomatic as in symptomatic children.[5,14–17] In a previous study at our institution that included non-ill patients 2–36 months of age having ambulatory surgery, HCoVs were detected in 2%,[18] lower than the 5% combined incidence in this study. The relationship between the coronaviruses detected and clinical manifestations is not proven to be causal. However, the illnesses occurred in the absence of other bacterial causes revealed by standard evaluations, which are admittedly imperfect. Second, our study was confined to a consecutive 13 months of study period and it is possible that variant viruses with different clinical manifestations circulate during other years. Third, we did not evaluate the serologic status for HCoVs; this may have been helpful in evaluating the clinical significance of coronavirus detection and also may have provided an estimate on how many coronavirus infections were not detected by polymerase chain reaction. Fourth, the sensitivity and specificity of the FilmArray assay used in this study are not well-defined. Limits of detection defined by the manufacturer are: HCoV-229E, 4 tissue culture infective dose 50 units per mL; HCoV-HKU1, 1.9×10^6 RNA copies per mL; HCoV-NL63, 5 tissue culture infective dose 50 units per mL and HCoV-OC43, 600 tissue culture infective dose 50 units per mL.[19] The specificity defined by the manufacturer in the prelicensure prospective clinical trial was > 99% for each of the 4 common HCoVs.[20] Finally, it is hard to generalize these data to a general pediatric population because we included only the patients who were hospitalized or visited the ED in of a tertiary referral hospital.

In conclusion, we observed a substantial burden of disease associated with both HCoV-OC43 and HCoV-NL63 in children <5 years of age. We plan to carry out further studies of the whole genomes of the HCoVs circulating during this season, which may allow us to understand the relationship between the virus circulating in St. Louis and previously described strains of HCoV-OC43, as well as the relationship of the viral genomes to clinical manifestations, and to understand the evolution of the viruses.

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